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Polybrominated Biphenyl (PBB) in the Growing Pig Diet

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Twelve pigs which averaged 13.7 kg were randomly allotted from litters to a corn-soybean meal grower diet containing 0, 20, or 200 ppm of polybrominated biphenyls (PPB). During a 16-week growth trial, average daily gain (kg), average daily feed (kg) and feed/gain for pigs on diets containing 0, 20, or 200 ppm of PBB, respectively, were 0.82, 2.45, 2.99; 0.67, 1.88, 2.79; 0.45, 1.23, 2.70. Mean daily gain differences between all lots were highly significant ($p < 0.01$). Blood from each pig was withdrawn biweekly through the first 8 weeks of the trial and at 4 week intervals thereafter. Hemoglobin and hematocrit differed significantly only at the 6 weeks bleeding, being reduced in pigs receiving 200 ppm of PBB. Erythrocyte reduced glutathione concentration and glutathione peroxidase activity were not significantly influenced by level of dietary PBB. Serum lactic dehydrogenase activity was significantly higher in control pigs than in either PBB supplemented lots at 16 weeks. There was no significant influence of PBB upon serum glutamic oxaloacetic transaminase, serum alkaline phosphatase or serum creatine phosphokinase. Based on these enzyme assays, PBB produced no evidence of significant necrosis of liver, myocardium, or skeletal muscle. There was no consistent effect of dietary PBB upon total serum protein concentration or electrophoretic profile. Pigs on either level of PBB did not have overt clinical signs of toxicity during the 16-week test period with the exception of a dermatosis on the ventral surface of two of the pigs receiving 200 ppm of PBB. There was a marked increase in liver weight of pigs receiving either level of dietary PBB. Heart, kidney, and adrenals of pigs receiving either level of dietary PBB were heavier as a percent of body weight than that of control pigs. Fat retention of PBB and urinary and fecal PBB excretion were significantly affected by dietary PBB level. Grossly, the glandular portion of the stomach appeared somewhat hyperplastic in pigs on 200 ppm of PBB. Two pigs which had received 200 ppm of PBB were placed on the control diet and over the next 14 weeks normal growth rate occurred. One of these pigs was killed and organ weights were normal. The other pig, a gilt, came into estrus. She was bred and conceived. At the end of gestation, four pigs were born. Three survived and grew normally; the one death at birth examined at gross necropsy did not reveal changes in organ size or other tissue alterations.

Introduction

Polybrominated biphenyls (PBB) are organic chemical compounds that are widely used for fire-resistant applications in both industrial and consumer products. In 1973, these chemical compounds were accidentally introduced into Michigan cattle feed. Since then many researchers have indicated a concern, not only about the effects of PBB on livestock performance, but also about PBB's potentially hazardous effects on human health.

Studies of PBB's effects have been reported on other species (*1–5*). However, information on the effect of PBB on swine is limited and further research is needed. The purpose of the present study was to evaluate the effect of different levels of PBB on performance of growing pigs and to determine if a high dietary PBB level during the growing phase of gilts precludes subsequent reproduction.

Materials and Methods

Twelve pigs averaging 13.7 kg were randomly allotted from litters into three groups of four pigs each fed a corn–soybean meal grower diet (Table 1) containing 0, 20, or 200 ppm of PBB. These diets, plus water, were offered *ad libitum*. The feeding trial

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Table 1. Composition of diets.

Ingredient	Control	PBB, 20 ppm	PBB, 200 ppm
Corn	79.0	79.0	79.0
Soybean meal 49	18.0	18.0	18.0
Limestone	1.0	1.0	1.0
Dicalcium phosphate	1.0	1.0	1.0
Salt	0.5	0.5	0.5
MSU VTM premix	0.5	0.5	0.5
PBB premix	–	1.0[a]	1.0[b]
PBB (by analysis) ppm	ND[c]	20.4	202.8

[a] PBB premix 0.2 wt. % BP-6 in corn oil. BP-6 is FireMaster FF-1, a mixture of brominated biphenyls, predominantly hexabromobiphenyl.

[b] PBB premix 2.0 wt. % BP-6 in corn oil.

[c] ND = not detectable; detection level of 0.01 ppm.

lasted for 16 weeks. All pigs were housed in a slotted floor pen about 1.5 m above ground level, and total excreta were collected and incinerated to destroy the PBB. Pig weights and feed consumption were recorded biweekly. Blood samples were collected from the anterior vena cava biweekly through the first 8 weeks and at 4-week intervals thereafter for hemoglobin (Hb) (*6*), hematocrit (Hct) (*7*), red blood cell reduced glutathione (RBC GSH) (*8*) concentration, and red blood cell glutathione peroxidase (RBC GPx) (*9*) activity determination. Serum samples were collected for alkaline phosphatase (AP) (*10*), glutamic-oxaloacetic transaminase (SGOT) (*11*), lactic dehydrogenase (LDH) (*12*), and creatine phosphokinase (CPK) (*13*) activity assay. Total serum protein concentration was determined by the Lowry method as modified by Miller (*14*), and the serum protein fraction profile was analyzed by agar gel electrophoresis (*15*). At week 4, urinary and fecal samples were collected and PBB concentrations were determined. Subcutaneous adipose tissue was also obtained by biopsy from the back immediately over the shoulder for PBB analyses by the GLC method (*16*). At termination, all pigs but two at the 200 ppm level were killed for gross necropsy examination. Selected organs were weighed, and samples of muscle, liver, kidneys, and subcutaneous and visceral adipose tissue were taken for PBB determination to characterize the pattern of PBB retention in growing pigs.

Results and Discussion

The effects of dietary treatment in pigs during a 16-week feeding trial on weight gain and feed consumption are shown in Table 2. Average daily gain and feed intake were significantly ($p < 0.01$) decreased by dietary PBB levels. The differences between pigs on 20 ppm and 200 ppm of PBB diets

Table 2. Performance of pigs in PBB study.[a]

	Dietary level of PBB		
	0 ppm	20 ppm	200 ppm
No. of pigs	4	4	4
Initial weight, kg	13.6	14.1	13.4
Final weight, kg	105.9[b]	92.2[c]	71.4[d]
ADG, kg	0.82[b]	0.67[c]	0.45[d]
ADF, kg	2.45	1.88	1.23
F/G	2.99	2.79	2.70

[a] Means with different superscripts differ significantly ($p < 0.01$).

were also significant ($p < 0.01$). It would appear that the higher the dietary PBB level, the greater the effect on feed intake and weight gain. It is also interesting to note that efficiency of feed conversion to gain was better for pigs on PBB treatments than for the controls. The effect of dietary PBB levels on weight gain was probably the result of reduced feed intake. This finding has been confirmed in chickens and Japanese quail as revealed in paired-feeding studies (*4*).

The average biweekly weight gain response to dietary levels of PBB expressed as growth curves is shown in Figure 1. Pigs receiving the 200 ppm PBB

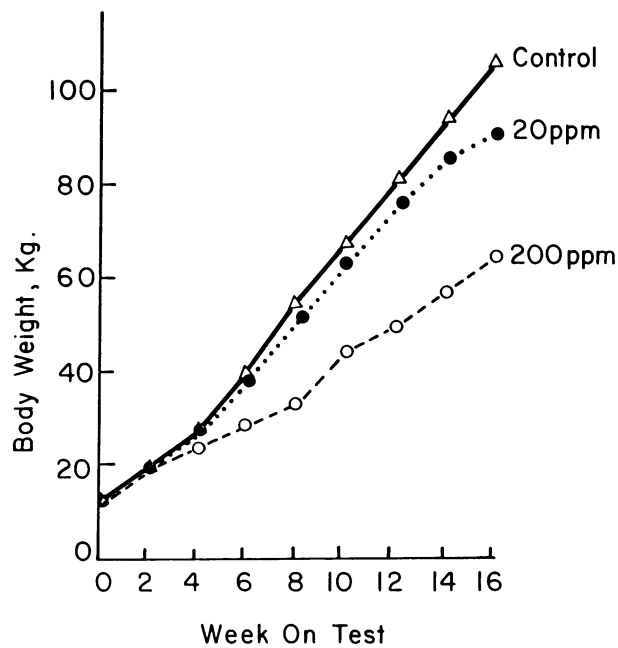

FIGURE 1. Effect of dietary PBB on growth.

diet grew significantly slower than the controls, and the difference increased as time progressed. Addition of 20 ppm of PBB to the diet did not significantly change the rate of growth until week 6 as compared to the controls. Three pigs, one from each treatment, are shown in Figure 2. As can be seen, there was no noticeable difference in their ap-

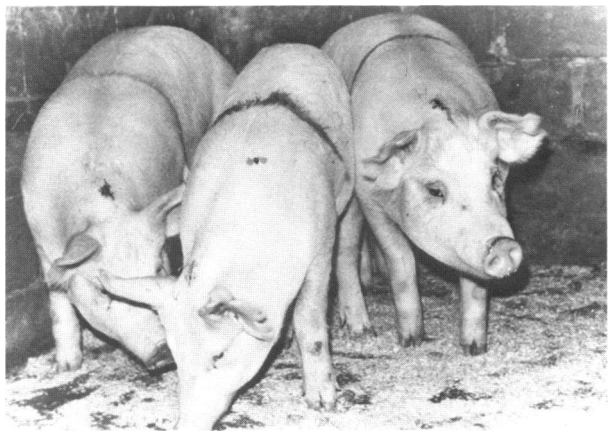

FIGURE 2. Pigs after 16 weeks on different dietary PBB levels. Control pig is on right, pig receiving 20 ppm is on left and pig receiving 200 ppm is in center. Other than growth depression, pigs receiving PBB appeared normal.

pearance. The pig on the right side was fed the control diet, the one on the left side, the 20 ppm PBB diet and the pig in the center, the 200 ppm PBB diet. Likewise, pigs on either level of PBB did not have overt clinical signs of toxicity during the 16-week trial, with the exception of a dermatosis on the ventral surface of two of the pigs receiving 200 ppm of PBB.

Hematological data, RBC GSH concentrations and RBC GPx activities are summarized in Table 3.

Table 3. Effect of dietary PBB levels on hemoglobin, hematocrit, RBC GSH, and RBC GPx activity.

	Dietary level of PBB,		
	0	20 ppm	200 ppm
Hemoglobin, g/dl			
2 weeks	10.7	11.1	11.4
4 weeks	12.1	11.4	11.2
6 weeks	12.6	12.7	11.4[a]
8 weeks	13.1	13.3	11.9
12 weeks	13.1	12.7	12.3
16 weeks	15.5	15.8	14.1
Hematocrit, %			
2 weeks	32.6	33.1	34.9
4 weeks	34.9	33.3	33.7
6 weeks	37.2	37.9	33.9[a]
8 weeks	38.4	37.4	34.3
12 weeks	39.6	38.3	37.5
16 weeks	39.5	38.5	39.5
RBC GSH, μmole/ml RBC[b]			
8 weeks	1.39	1.51	1.53
12 weeks	1.36	1.40	1.41
16 weeks	1.32	0.97	1.23
RBC GPx, units/mg Hb[c]			
8 weeks	15.2	12.9	13.6
12 weeks	14.2	11.7	16.0
16 weeks	16.4	13.5	13.0

[a] Significantly lower than other treatments ($p < 0.05$).
[b] RBC GSH = Red blood cell reduced glutathione.
[c] RBC GPx = Red blood cell glutathione peroxidase.

Blood Hb concentration and Hct values were not significantly different between the 20 ppm group and controls. However, Hb and Hct values tended to be lower in pigs on the 200 ppm level, although differences were significant only at week 6. PBB has been shown to depress Hb and Hct values in the chicken (*4*). RBC GSH concentrations and RBC GPx activities were not significantly affected by supplemental PBB, although addition of either level of PBB tended to increase RBC GSH concentration at week 8 and 12, and then decreased it at week 16. On the other hand, with addition of PBB to the diets the activity of RBC GPx tended to decrease. A possible explanation for these differences may be the refusal of food. Decreased RBC GSH concentrations and reduced RBC GPx activities have been observed in fasted animals (*17, 18*).

Total serum protein concentration and serum protein fractionation profiles are presented in Table 4. There was no consistent effect of dietary PBB upon total serum protein concentration or electrophoretic profile.

The effect of dietary PBB levels on the activities of serum AP, GOT, LDH, and CPK are presented in Table 5. No significant influence of dietary PBB levels on these serum enzyme activities was observed, except that serum LDH activity was significantly higher in control pigs than in either PBB supplemented lots at week 16. Based on these enzyme assays, growing pigs fed either 20 or 200 ppm of PBB for 16 weeks showed no evidence of significant necrosis of liver, myocardium or skeletal muscle. It was confirmed later by gross examination at necropsy that no severe lesions due to PBB were present except that the glandular portion of the stomach appeared somewhat hyperplastic in pigs on 200 ppm of PBB.

A summary of the effects of dietary PBB levels upon selected organ weights is presented in Table 6. The PBB supplement at either level significantly increased the absolute weight of liver and kidneys. The effect on heart was somewhat variable. When organ weights were expressed as a percentage of body weight, variations were observed. Liver, kidneys, and heart were a greater percentage of body weight in PBB-fed pigs. Adrenals tended to increase in weight either on 20 or 200 ppm levels. These alterations in organ or tissue size have also been observed in chickens, Japanese quail and rats (*1, 3, 4, 19*). This is probably a normal detoxification response. It is similar to the known effects of a normal liver response to the presence of some known drugs (e.g., phenobarbitals) where the animal body attempts to detoxify potentially harmful chemicals. Based on these observations, it seems that the liver responds sensitively to PBB in the diet by increas-

Table 4. Effect of dietary PBB levels on serum protein concentration and serum protein fraction profile.

Weeks on test	PBB level, ppm	Protein, FireMg/100 ml	Albumin, %	α-Globulin, %	β-Globulin %	γ-Globulin, %
	0	6.63	32.4	18.2	21.1	28.4
2	20	6.69	29.2	23.8	21.9	25.0
	200	6.67	34.4	24.4	19.7	23.0
	0	6.88	37.5	17.8	26.1	24.6
4	20	6.65	35.5	22.7	22.0	19.8
	200	6.43	39.9	22.2	19.1	18.0
	0	7.53	41.2	16.9[b]	22.4	19.6
6	20	7.63	42.7	18.6[ab]	20.3	18.3
	200	7.25	41.4	22.0[a]	20.0	16.6
	0	7.68	39.0	18.4	22.9	19.7
8	20	8.03	37.2	21.0	22.9	19.0
	200	7.31	36.8	21.6	21.9	19.8
	0	7.64	39.1	18.7	22.7	14.6
12	20	7.85	39.0	20.7	22.6	17.5
	200	7.18	39.0	20.6	21.1	16.3
	0	8.00	42.9	18.8	22.2	16.1
16	20	8.60	38.0	22.7	21.4	18.0
	200	7.62	34.8	23.1	22.1	15.0

[a,b] Means with different superscripts differ significantly ($p < 0.05$).

Table 5. Effect of dietary PBB levels on the activity of serum AP, GOT, LDH and CPK.[a]

	Dietary level of PBB		
	0	20 ppm	200 ppm
AP, sigma units/ml			
2 weeks	3.65	4.41	2.95
4 weeks	4.88	4.43	3.32
6 weeks	5.14	5.42	3.57
8 weeks	5.87	4.97	4.41
12 weeks	3.69	4.20	2.64
16 weeks	3.28	2.25	2.81
GOT, sigma-Frankel units/ml			
2 weeks	50.3	38.8	47.6
4 weeks	42.9	38.1	39.0
6 weeks	42.3	47.7	39.6
8 weeks	33.4	33.5	31.9
12 weeks	26.5	30.4	29.4
16 weeks	20.3	20.3	22.7
LDH, Berger-Broida units/ml			
2 weeks	663.8	770.1	835.5
4 weeks	740.0	711.9	598.4
6 weeks	809.6	857.0	831.0
8 weeks	823.6	827.9	790.6
12 weeks	676.1	835.6	595.0
16 weeks	615.6[b]	481.9	497.4
CPK, sigma units/ml			
2 weeks	25.4	18.2	17.9
4 weeks	21.4	40.9	9.0
6 weeks	39.7	55.3	32.8
8 weeks	13.9	48.4	34.0
12 weeks	25.6	68.5	17.3
16 weeks	17.7	46.7	21.0

[a] AP = alkaline phosphatase, GOT = glutamic-oxaloacetic transaminase, LDH = lactic dehydrogenase, CPK = creatine phosphokinase.
[b] Significantly ($p < 0.05$) greater than other values.

Table 6. Effect of dietary PBB levels on organ weights or percentage of body weight.

	Dietary level of PBB[a,b]		
Organ	0	20 ppm	200 ppm
Liver, g (%)	1346[a] (1.27)[c]	1785[d] (1.94)[d]	3038[e] (4.25)[e]
Kidneys, g (%)	285.5[c] (0.27)[c]	360.6[d] (0.39)[d]	310.5[c] (0.43)[d]
Heart, g (%)	292.2[c,d] (0.28)[c]	331.1[a] (0.36)[d]	271.5[d] (0.38)[d]
Spleen, g (%)	149.0 (0.14)	155.0 (0.17)	130.0 (0.19)
Thyroid, g (%)	9.88 (0.009)	8.90 (0.010)	9.05 (0.013)
Adrenals, g (%)	6.20 (0.006)[c]	7.18 (0.008)[c,d]	6.50 (0.009)[d]

[a] Numbers in parentheses are percent of body weight values.
[b] Means with different superscripts differ significantly ($p < 0.05$).

ing its functional mass. The effects of dietary treatment on PBB retention and excretion patterns at week 4 and at termination are combined in Table 7. Fecal excretion is a major excretory route for PBB. Dietary PBB levels had a significant effect on PBB retention in adipose tissue. These data suggest that the characteristics of PBB retention are similar to those of other halogenated organic compounds (e.g., PCB, DDT) in biological tissues (*20*).

It would appear that adipose tissue PBB levels are a sensitive reflection of the concentration of PBB in the diet. On the 20 ppm level, tissue PBB residue in muscle was lower than in liver, and the kidneys had the lowest concentration. On the 200 ppm level, the PBB residue was similar between muscle and liver and slightly lower in kidneys. The highest level of residue was found in adipose tissue, and it is apparent that the extent of accumulation of PBB in fat was directly proportional to the length of

Table 7. Effects of dietary PBB levels on tissue PBB retention and PBB excretion at week 4 and week 16.[a]

	Dietary levels of PBB[b]		
	0	20 ppm	200 ppm
Week 4			
Back fat, ppm	ND (4)	0.33 (4)	6.73[c] (4)
Urine, ppm	ND	0.015	0.07
Feces, ppm	ND	21.3	182.0
Week 16			
Muscle, ppm	ND (4)	6.60 (4)	18.37[d] (2)
Liver, ppm	ND (4)	8.46 (4)	17.19[c] (2)
Kidneys, ppm	ND (4)	0.93 (4)	13.52[c] (2)
Backfat, ppm	ND (4)	64.04 (4)	502.79 (2)
Leaf fat, ppm	ND (4)	42.93 (4)	459.08[c] (2)

[a] All tissue values expressed on a wet basis; fecal samples were oven-dried and ground; urine was preserved with 6 *N* HCl. ND = not detectable at detection limit of 0.01 ppm.
[b] Numbers in parentheses are the numbers of observations.
[c] Significantly greater than other values ($p < 0.01$)
[d] Significantly greater than other values ($p < 0.05$).

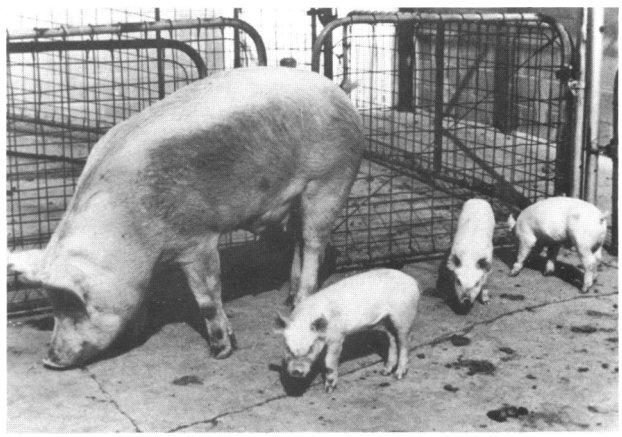

FIGURE 3. This gilt received 200 ppm dietary PBB for 16 weeks, then was placed on control diet for 14 weeks, was bred, she conceived, farrowed and nursed a litter of 4 pigs, three of which were alive and well at weaning (5 weeks).

exposure. Since no PBB withdrawal study was conducted, a biological half-life of PBB in tissue could not be calculated.

Table 8 provides information on one pig which was originally fed 200 ppm PBB and then was returned to a normal diet. The time on the normal diet was 102 days; the average daily gain was 0.70 kg. Some organ weights of this pig expressed on both an absolute basis and as a percent of body weight are included in this table. Comparisons with the organ weights (as percent of body weight) obtained from pigs on the control diet and pigs studied by McMeekan (*21*) suggest that after PBB withdrawal, organ weights returned to a normal range.

Figure 3 shows a gilt which was one of the pigs originally on 200 ppm of PBB. This gilt was returned to a normal diet for 14 weeks, grew at a normal rate, exhibited estrus, was bred and conceived. Four pigs

Table 8. Organ weights and performance of pig switched from PBB (200 ppm) diet to normal grower diet.

	PBB withdrawal	Control	Literature[a]
Initial weight, kg	65.0	13.6	–
Final weight, kg	136.4	105.9	100
Days on test	102	112	–
ADG, kg	0.70	0.82	–
Liver, g (%)	2440 (1.8)	1346 (1.3)	1745 (1.75)
Heart, g (%)	335.2 (0.25)	292.2 (0.28)	266 (0.27)
Kidneys, g (%)	379.6 (0.28)	285.5 (0.27)	225 (0.23)

[a] Data of McMeekan (*21*).

were born at the end of the gestation period; three survived, grew normally, and showed no gross external abnormalities. They are shown in Figure 3 with their mother. Whether the one death at birth was related to PBB is not clear. The gross necropsy examination did not reveal changes in organ size or other tissue alterations. Hb and Hct values obtained from the sow and baby pigs were normal as compared to those reported by Miller et al. (*22*). Compared with the data of controls and others (Ku, unpublished data), total serum protein and the enzyme activities of serum AP, GOT, CPK, and LDH from the sow and baby pigs were also in the normal range, as were RBC GSH concentrations and RBC GPx activities (Parsons and Brady, unpublished data).

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